Received: 30 July 2014 Revised: 15 September 2014 Accepted article published: 20 October 2014 Published online in Wiley Online Library: 7 November 2014

(wileyonlinelibrary.com) DOI 10.1002/pi.4822

Preparation of polymeric nanoparticles by novel electrospray nanoprecipitation

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Abstract

Polymeric nanoparticles have important applications in drug delivery, biotechnology, diagnostics and energy harvesting. We report a new technique named electrospray nanoprecipitation, which combines electrospray with agitated solvent displacement. The process enables one-step formation of polymeric nanoparticles <100 nm in size that are near-monodisperse with a diameter range significantly lower than could be obtained using either electrospray or agitated solvent displacement technique alone. Both reduction of polymer solution concentration and the addition of poly(vinyl alcohol) emulsifier in the water–non-solvent medium further reduce the average particle diameter. The technique provides an effective and straightforward method to further reduce the size range of near-monodisperse nanoparticles achievable in a single step, which can be readily adapted for reducing the achievable size range of core–shell structures using popular one-step encapsulation techniques such as coaxial electrospray.

Keywords: electrospray; nanoprecipitation; nanoparticles; polymethylsilsesquioxane

INTRODUCTION

Nanotechnology refers to the design, characterisation, production and application of materials at atomic, molecular and macromolecular scales. More than fifty years ago, Richard Feynman[1] proposed the direct manipulation of individual atoms as a powerful mode of synthetic chemistry, giving rise to 'nanotechnology', a term later coined by Norio Taniguchi[2] in 1974 to describe the bottom-up formation of materials from atoms and molecules at a tolerance of one billionth of a metre. Advances in materials science and processing technologies over the last 25 years expanded the concept of nanotechnology to include top-down transformation of macroscopic materials into nanomaterials.[3,4]

Polymeric nanoparticles (NPs) have important applications in drug delivery,[5,6] biotechnology,[7] diagnostics,[8,9] and energy harvesting.[10] NPs can be made via bottom-up or top-down methods, or a combination of both.[5,11] Bottom-up methods involve synthesising the NPs through particle nucleation during polymerisation reactions of monomeric/oligomeric precursors, such as emulsion polymerisation and interfacial polycondensation. These techniques often require additional strategies and processing stages to remove residue precursors, reagents and surfactants, and they cannot be applied to preformed materials or reconstituted natural materials. On the other hand, top-down processing reduces macroscopic materials such as preformed or natural polymers in solutions into NPs. Examples include salting-out/emulsification–diffusion,[12–14] supercritical fluid techniques,[15] electrospray[16,17] and solvent displacement.[5,11,18,19] Unlike emulsification and supercritical fluid methods that require high-energy input devices like sonicators and high-cost devices such as high-pressure homogenisers, solvent displacement and electrospray are straightforward and versatile techniques featuring advantages like ambient conditions, single-step processing, high reproducibility, high yield and economical set-up.[17,19]

Electrospray stretches and breaks up a leaky dielectric liquid from a macroscopic mass (millimetre range) to ultrafine droplets in the nanometre/micrometre range. Energy is supplied by means of electrohydrodynamic forces to overcome the surface tension of the liquid.[20,21] Specifically, a leaky dielectric, e.g. a dilute polymer solution, is infused in a needle electrode connected to a kilovolt-applied voltage. Inside a strong electric field, charge builds up on the polymer solution. At a range of applied voltages and infusion rates, the electrostatic force overcomes the surface tension of the liquid and the meniscus of the liquid held at the needle exit deforms into a conical shape.[22] A fine jet emanates from the apex of the cone and accelerates towards a grounded electrode, breaking up into fine NPs along the jet path due to Rayleigh–Plateau instability.[23] A stable cone-jet, governed by

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electromechanical principles, is the most desirable electrospray condition for near-monodisperse polymeric NP generation.[24]

Solvent displacement is a common process in chemistry and pharmaceutics. A polymer solution is mixed with a non-solvent that is miscible with the solvent of the polymer. Phase separation occurs. Particle formation during solvent displacement involves particle nucleation, molecular growth and particle agglomeration. Controlling the rate of mixing, the relative concentrations of the solvent/polymer/non-solvent, the temperature and the pressure allows controlled phase separation, yielding polymeric NPs of various size ranges.[25,26]

The key phenomenon in solvent displacement is nucleation, driven by the local super-saturation and the interfacial tension between the solid particles and the liquid medium.[25] Low nucleation rates correspond to low rates of local super-saturation, leading to fewer and larger particles; whereas high nucleation rates result in a larger population of smaller particles. Super-saturation occurs when the polymer solution contains more solute than the solvent can dissolve under equilibrium conditions. This is intrinsically dependent on the solid–liquid contact, where rapid molecular exchange occurs between the solvent and the non-solvent surrounding the polymeric macromolecules. Mechanical agitation or stirring is the most frequently used method in chemical process industries to achieve high solid–liquid contact.[27–29]

Production of NPs by solvent displacement alone is in some cases also known as nanoprecipitation.[10] The technique is challenged by the formulation of a drug–polymer–solvent–non-solvent system that allows efficient drug encapsulation in the NPs, because the solvent and the non-solvent of the polymer are miscible.[5] Electrospray techniques do not suffer this drawback.[16,23] Combining the two methods appears as a logical approach. Electrospray has frequently reported jetting NPs into a static non-solvent medium (wet electrospray), to provide shock absorption during particle collection.[9] However, NP formation via mechanically agitated precipitation of electrosprayed particles has not been investigated. This communication describes electrospray nanoprecipitation, a one-stage preparation method for forming polymeric NPs that combines electrospray and agitated solvent displacement. The resultant polymeric NPs are near-monodisperse with diameters significantly lower than those obtained using either technique alone.

Polymethylsilsesquioxane (PMSQ) was used as a model polymer. PMSQ can be directly converted to ceramic products via pyrolysis.[30,31] An interesting polymer for materials engineering of both polymeric and ceramic products, PMSQ is non-polar, biocompatible with a ladder-like structure, has good heat resistance, good slip properties and disperses excellently into organic liquids, making the polymer a suitable choice for both theoretical studies and a variety of applications.[31] The pyrolysed ceramic silicon oxycarbide has high thermal and chemical durability, low density, low dielectric constant, antireflection properties, biocompatibility and high mechanical strength. These properties make PMSQ an attractive material for applications in medicine and biomedical engineering.[32,33]

MATERIALS AND METHODS

Materials

PMSQ (average molecular weight (M_w) = 4300 g mol^{-1}, polydispersity = 3.6) was obtained from Wacker Chemie AG. Ethanol and poly(vinyl alcohol) (PVA; 87–89% hydrolysed, M_w = 13 000–23 000 g mol^{-1}) were obtained from Sigma-Aldrich, Poole, UK. All materials obtained were of analytical grade and used as received. PMSQ solutions (10% w/w) were made by dissolving an appropriate amount of PMSQ in ethanol. PVA in water (1 and 5% w/w) was also prepared.

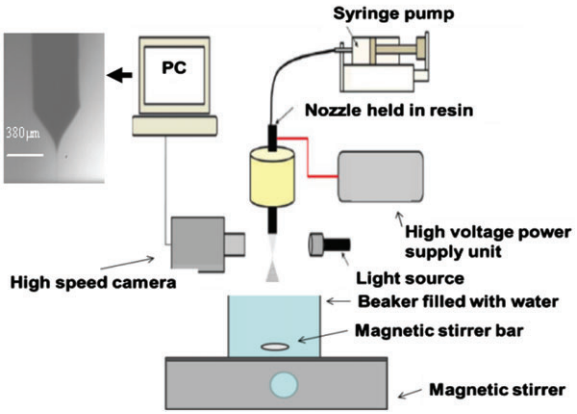

Figure 1. Schematic representation of the electrospray nanoprecipitation set-up. A high-speed camera image of the stable cone-jet is also included.

Experimental set-up

The experimental set-up for electrospray nanoprecipitation is illustrated in Fig. 1. A high-speed camera image of the stable cone-jet in electrospray is also included in Fig. 1. Specifically, the polymer solution was introduced to a nozzle of 330 µm inner diameter, at a constant flow rate of 15 μL min^{-1} using a Harvard syringe pump (Harvard Apparatus Ltd, Edenbridge, UK). The nozzle was connected to the positive electrode of a high-voltage supply (Glassman Europe Ltd, Bramley, UK). Distilled and deionised water, a non-solvent of PMSQ, miscible with ethanol, was used as the collection medium. Electrosprayed droplets were collected by placing a beaker containing 80 mL of water stirred by a magnetic stirrer (20 mm) at a constant rotary speed of 1000 rpm (using a magnetic hotplate, Kika-Werke, Germany). The beaker was placed on a grounded stainless steel laboratory jack with a deposition distance of 50 mm between the charged nozzle exit and the collecting water surface. Experiments were conducted under conditions of atmospheric pressure, relative humidity ranging between 40 and 55% and ambient temperature of 20–22 °C. The operating applied voltage was optimised to maintain a stable cone-jet over the duration of particle collection. This was at 8.0 ± 1.5 and 5.0 ± 0.2 kV, respectively, for 10 and 5% (w/w) PMSQ solutions.

Three control experiments forming particles by wet electrospray, dry electrospray and agitated solvent displacement were carried out to compare with the particles produced in electrospray nanoprecipitation. In solvent displacement, the polymer solution was infused at 15 μL min^{-1}, and progressively dripped from a nozzle of 330 µm inner diameter into 80 mL of water. The non-solvent water was in a 100 mL glass beaker, mechanically stirred by a magnetic stirrer (20 mm length) at a constant rotary speed of 1000 rpm. Particles were collected on both dry aluminium and in a beaker of 80 mL of water without stirring in control experiments using dry and wet electrospray, respectively.

Particle characterisation

Particle morphology was characterised using an optical microscope (Nikon Eclipse, model ME600) and a field emission SEM instrument (model JSM-6301 F). For SEM imaging, dry samples were obtained by drying under ambient conditions a drop of the collected particle suspension on aluminium foil. Subsequently

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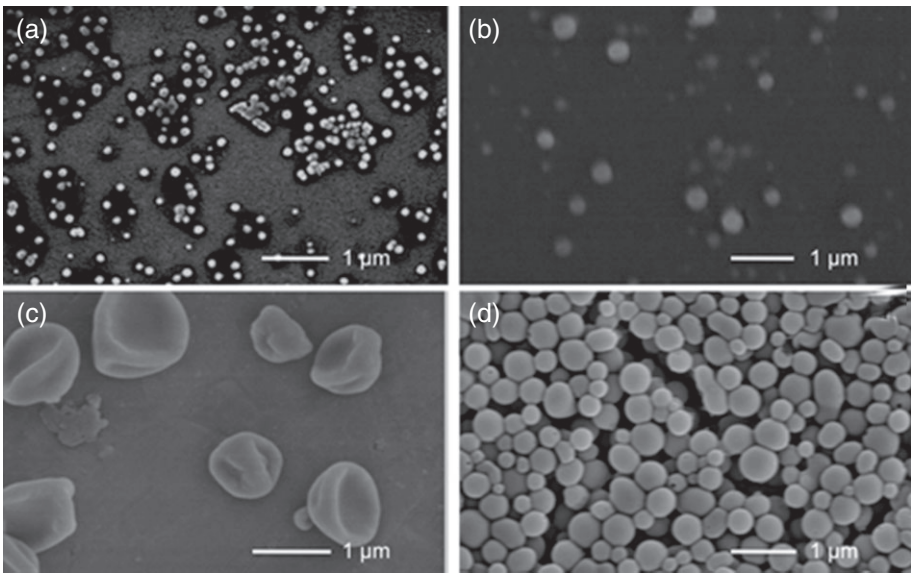

Figure 2. SEM images of PMSQ particles produced using (a) electrospray nanoprecipitation, (b) agitated solvent displacement, (c) dry electrospray and (d) wet electrospray. In all processes, a 10% (w/w) PMSQ solution in ethanol was infused at 15 μL min^{-1}, exiting a nozzle of 330 μm inner diameter, and collected at a distance of 50 mm. Variations in the processes: (a) particles produced at 9 kV over 50 mm from the nozzle to the surface of 80 mL of water in a 100 mL beaker, mechanically stirred at 1000 rpm; (b) particles produced without an applied potential but otherwise under the same conditions as in (a); (c) particles produced at 9 kV over 50 mm to a grounded piece of dry aluminium foil, i.e. the solvent dried in air; (d) particles produced under the same conditions as in (a) but without mechanical stirring.

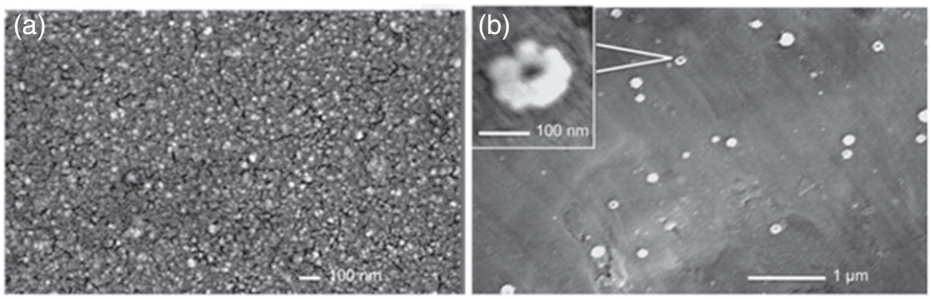

Figure 3. SEM images of PMSQ particles from 5% (w/w) PMSQ solutions, produced using (a) electrospray nanoprecipitation and (b) agitated solvent displacement without electrospray.

each sample was coated with gold using a sputtering machine (Edwards S 1 50B sputter coater) for 120 s prior to SEM observation at an accelerating voltage of 3–5 kV. The diameters and the size distribution profiles of the particles were characterised based on 100 measurements, using ImageJ, a public-domain Java image-processing program.

RESULTS AND DISCUSSION

Conventional electrospray achieves particle solidification by static solvent displacement (wet electrospray). The droplets under a strong electric field are sprayed directly into a static non-solvent liquid medium such as water or ethanol. Near-monodisperse polymeric particles with diameters from 100 nm to 10 μm can be prepared in this way.[34] However, a one-step preparation of surfactant-free polymeric NPs of 100 nm in diameter has been a challenge.[5,35]

The new NP-forming process of electrospray nanoprecipitation overcomes the challenge mentioned above with a set-up modification (stirring the liquid collector, Fig. 1). Figure 2 shows a comparison of the diameter and the degree of aggregation of NPs produced using the various processes discussed here: electrospray nanoprecipitation (Fig. 2(a)), agitated solvent displacement without electrospray or surfactants (Fig. 2(b)) and conventional electrospray processes with particles collected either on dry aluminium foil (Fig. 2(c)) or by static solvent displacement (Fig. 2(d)).

Electrospray nanoprecipitation consistently generates near-monodisperse surfactant-free NPs, with an average particle diameter of 62 ± 20 nm (Figs 2(a) and 3). Solvent displacement can achieve monodisperse polymeric NPs with a size range of approximately 50–300 nm.[5,19,36] In this work, NPs with an average diameter of 206 ± 71 nm are produced using the agitated solvent displacement technique (Fig. 2(b)). The process progressively adds PMSQ dissolved in ethanol from a nozzle of 330 μm inner orifice diameter to 80 mL of water under stirring. In contrast, dry electrospray and wet electrospray techniques produce PMSQ particles of 922 ± 153 nm (Fig. 2(c)) and 333 ± 69 nm (Fig. 2(d)), respectively.

During solvent displacement, fluid dynamics and rate of mixing determine the degree of solid–liquid contact and the local value of super-saturation, which consequently influence the rate of nucleation and particle size.[25,28] Mechanical agitation or stirring is the most frequently used method in chemical process industries to achieve high solid–liquid contact.[27–29] It is therefore logical that electrospray nanoprecipitation produces smaller particles

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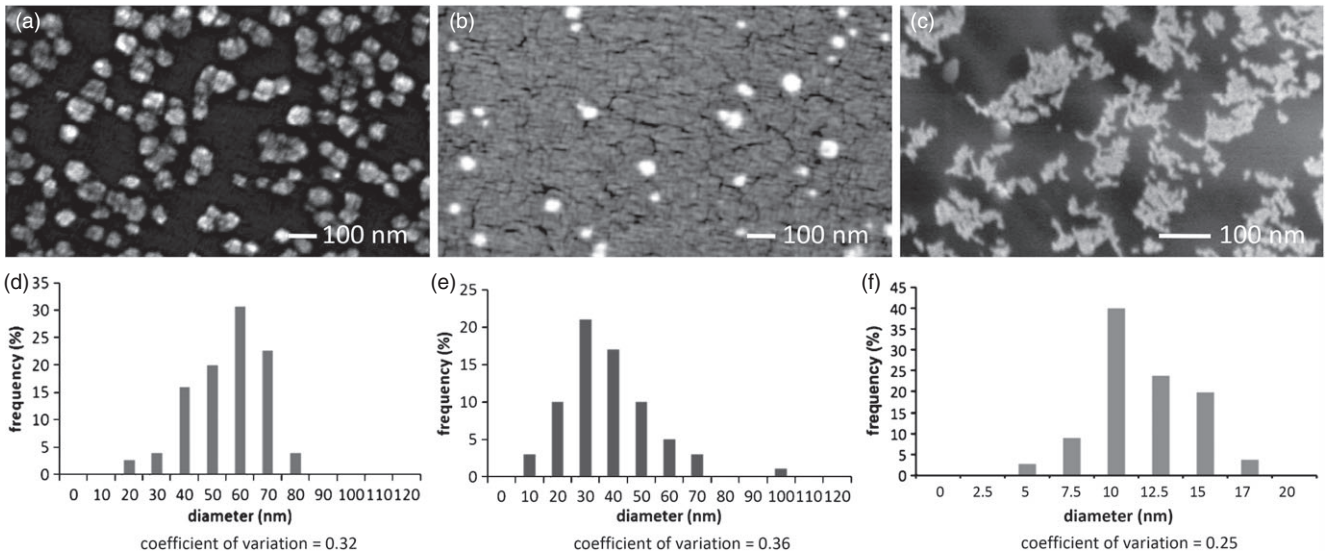

Figure 4. SEM images of PMSQ NPs from 10% (w/w) PMSQ solutions, produced using electrospray nanoprecipitation with various concentrations of PVA added to the particle-precipitating water medium: (a) 0% PVA; (b) 1% (w/w) PVA; (c) 5% (w/w) PVA. Particle size decreases due to reduced secondary aggregation while the particles remain suspended in water. (d–f) Corresponding size distribution profiles. The coefficient of variation was calculated by standard deviation/mean.

compared to the static wet electrospray technique. Hence, the rate of particle formation and rate of mixing have competing timescales. Up to a critical value, increasing rate of mixing reduces the particle size.[25] It is known that the initial polymer concentration affects the rate of particle formation.[25] When the PMSQ polymer solution concentration is reduced from 10 to 5% (w/w), NP diameter decreases from 62 ± 20 to 20 ± 7 nm (compare Figs 2(a) and 3(a)).

Higher resolution images of 5% PMSQ particles produced using agitated solvent displacement without electrospray (inset of Fig. 3(b)) show that the particles are composed of a ring assembly of aggregating, smaller particles. Aggregation is considered a secondary process because its rate is proportional to the second power of the number density (i.e. concentration) of particles.[25] The proportionality constant is the aggregation frequency, which depends on the size of the particles and on the mechanism of collision, predominantly perikinetic aggregation through Brownian motions.[25]

To reduce aggregation and stabilise particles in suspension, emulsifiers are commonly used and PVA is the most common NP-stabilising emulsifier in emulsification–diffusion techniques.[11] We added various concentrations of PVA in the non-solvent medium, water, to investigate the effect of the surfactant on particle aggregation in the liquid medium after particle production by electrospray nanoprecipitation of 10% (w/w) PMSQ solutions. The average particle size observed using SEM reduces from 62 ± 20 nm in the absence of PVA (Fig. 4(a)), to 42 ± 15 nm with 1% (w/w) PVA (Fig. 4(b)), to very fine particles with an estimated diameter range of 10–20 nm with 5% (w/w) PVA (Fig. 4(c)).

CONCLUSIONS

We report a new technique called electrospray nanoprecipitation, which combines electrospray with agitated solvent displacement. The process enabled one-stage formation of polymeric NPs <100 nm in size that were near-monodisperse with a diameter range significantly lower than that obtained using either electrospray or agitated solvent displacement technique alone. Both reduction of polymer solution concentration and the addition of PVA surfactant in the water non-solvent medium further reduced the average particle diameter. The technique provides an effective and economical method to further reduce the size range of near-monodisperse NPs achievable in a single step. The process is also useful for reducing the achievable size range of core–shell structures using popular encapsulation techniques such as coaxial electrospray.

ACKNOWLEDGEMENTS

The authors thank the Japan Patent Office for funding a sabbatical period of training and research for TO that helped initiate this work. CJL and ME thank EPSRC and a UCL Knowledge Transfer & Enterprise Award for supporting their research. We also thank the Archaeology Department (UCL) for use of their microscope facilities.

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